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# Intra-tree Activity of Male Mediterranean Fruit Flies (Diptera: Tephritidae): Effects of Posteclosion Light, Crowding, Adult Diet, and Irradiation

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**ABSTRACT** Laboratory-reared Mediterranean fruit flies *Ceratitis capitata* (Wiedemann) were held under varying conditions of light, density, food, and irradiation prior to release of males on potted guava, *Psidium guajava* L., plants in outdoor cages. Male activity after release was measured in terms of number of leaves visited and duration of flights within the plant canopy. Males held continuously in the dark were more active than those held under a photoperiod of 12:12 (L:D). Males held continuously in the dark at high densities were more active than those held continuously in the dark at low densities. Males provided sugar and protein were more active than those provided only sugar. Radiation did not appear to affect activity of flies tested. Implications of these findings to sterile insect programs are discussed.

Sterile Insect Technique (SIT) is an important control method for Mediterranean fruit fly, *Ceratitis capitata* (Wiedemann), that involves mass production, sterilization, and release of millions of flies into wild populations (Knipling 1955, Steiner *et al.* 1962, Harris *et al.* 1986, Wong *et al.* 1986, Vargas *et al.* 1994). The manager of a sterile release program needs to know about the behavior of released flies for a successful program (Baker & Chan 1991b, Plant & Cunningham 1991). Baker & Chan (1991a) suggested that the distribution of released flies among trees is due to genetically determined flight times modified by hunger, sex, and environmental factors. Foraging is an important resource finding behavior that affects the distribution of flies (Prokopy & Roitberg 1989), both wild and released. Baker and van der Valk (1992) observed that sterile Mediterranean fruit flies disappeared rapidly after release. Males remaining in the tree canopy formed small aggregations. Similarly, Vargas *et al.* (1995) observed sharp declines in sterile male numbers within 5 d of aerial release over coffee (*Coffea arabica* Linnaeus) fields.

Sterile Mediterranean fruit flies are irradiated as pupae and placed inside screened emergence boxes (40,000 per box). Newly emerged adults are provided water in agar, fed sugar, and held in darkness for 3 d before release (Cunningham *et al.* 1980, California Department of Food and Agriculture 1987, Vargas *et al.* 1994, 1995). Little is known about the effects of these holding conditions on the ability of released sterile male flies to adapt to their surroundings and forage. The objective of the present study was to examine the effects of pre-release holding conditions of light, food (sugar and/or protein), density, and irradiation on male activity shortly after release.

## MATERIALS AND METHODS

Studies were conducted at the Tropical Fruit and Vegetable Research Laboratory, USDA-ARS, Honolulu, Hawai'i, from March to December 1993. Mediterranean fruit flies used in this study were reared as described previously (Vargas 1989). This colony has been maintained for approximately 320 generations. Newly eclosed flies were maintained in screened wooden cubical cages (27 cm per side) at 25 ±3 °C and 60 ±10 %RH and provided with sugar and water for 3 d.

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**Table 1.** Foraging behavior (number of leaves visited, total time in flight, and total time in tree) of male Mediterranean fruit flies ($\bar{X} \pm SE$) held under different combinations of light, density food and irradiation

| Experiment | N | No. of leaves visited[a] | | Time in flight (sec)[a] | | Total time in tree (min)[b] | |
|---|---|---|---|---|---|---|---|
| Light Exposure | 25 | dark | light | dark | light | dark | light |
| | | 9.1 ± 2.8[a] | 2.8 ± 0.7[b] | 18.6 ± 5.3[a] | 6.6 ± 1.7[b] | 4.7 ± 1.0[a] | 2.9 ± 0.9[a] |
| Adult Density | 25 | high | low | high | low | high | low |
| | | 15.3 ± 3.3[a] | 5.9 ± 1.3[b] | 26.6 ± 5.7[a] | 10.6 ± 2.4[b] | 7.6 ± 1.1[a] | 7.0 ± 1.3[a] |
| Food Type | 25 | sugar | protein & sugar | sugar | protein & sugar | sugar | protein & sugar |
| (high density) | | 3.4 ± 2.6[b] | 6.7 ± 1.0[a] | 6.4 ± 1.2[b] | 15.2 ± 3.0[a] | 6.4 ± 1.2[a] | 4.3 ± 0.9[a] |
| Irradiation | 25 | normal | irradiated | normal | irradiated | normal | irradiated |
| Treatment | | 7.8 ± 1.4[a] | 8.3 ± 1.0[a] | 10.5 ± 1.8[a] | 10.1[a] ± 1.1[a] | 7.5 ± 1.2[a] | 7.5 ± 0.9[a] |

a. Paired values not followed by the same letter are not significantly different at the 0.05 level according to a t test.
b. Paired values not followed by the same letter are not significantly different at the 0.05 level according to a Mann-Whitney U test.

We conducted four experiments to determine the effects of: (1) light exposure, (2) adult density, (3) protein and (4) irradiation. To obtain light- and dark-exposed flies, pupae were held in two rooms with photoperiods of 12:12(L:D) h and 0:24(L:D) h, respectively. For all other tests, adults were held in the room maintained with a photoperiod of 0:24(L:D) h before testing. Where the effect of fly density was investigated, either 100 or 4 ml of pupae were allowed to eclose in separate cubical cages, which constituted high and low density conditions, respectively. Diets of adults were compared and consisted of a 3:1 volumetric mixture of sugar and enzymatic yeast hydrolysate (U.S. Biochemical, Cleveland, OH) or plain sugar. High densities of adults were maintained before testing for comparisons involving light exposure, food type and radiation treatment. The sugar-protein mixture served as the food source for comparisons involving light exposure, adult density and radiation treatment. Irradiated pupae received 15 Krads from a cobalt-60 source 2 d before eclosion for comparison with nonirradiated pupae.

All experiments were conducted between 0900–1500 h in one of 3 cylindrical (3 m high by 3 m diam.) clear-nylon-screen field cages (Chicopee Manufacturing Co., Gainesville, GA) containing two potted guava trees, *Psidium guajava* L., closely spaced near the center of the cage (Prokopy *et al.* 1993). These cages were covered with blue opaque plastic tarpaulins to protect the test arena from frequent rains. A single leaf, centrally located and at the base of the tree canopy served as a release site for males which were released and observed individually. Temperature within the tree canopy during these experiments ranged from 25–32 °C. To accommodate possible differential inter-treatment effects of temperature and time of day on male behavior, we alternated releases between the two treatments of an experiment. A trial was terminated when a fly left the tree canopy and alighted on the field cage or when 15 min expired. Data were recorded on number of leaves visited, duration of individual flights within the tree canopy, and total time spent in the tree canopy. Each comparison was replicated with 25 male flies. Data were analyzed statistically by a Student's t-test or the Mann-Whitney U test at the $P = 0.05$ level (SAS Institute 1987).

## RESULTS

Mediterranean fruit flies held continually in the dark visited significantly greater numbers of leaves and were in flight significantly longer than those held under light (Table 1). Flies held continually in the dark at high densities visited significantly more leaves and were in flight significantly longer than those held continually in the dark at low densities. Flies fed sugar and protein visited significantly more leaves and were in flight significantly longer than those fed only sugar. Radiation did not significantly affect the activity patterns of flies tested.

## DISCUSSION

Rapid disappearance of sterile Mediterranean fruit flies shortly after release has been a concern to sterile insect program managers (Baker & Chan 1991b, Shelly *et al.* 1993, Vargas *et al.* 1995). The fate of flies remains uncertain, but there are many possibilities. Flies may (1) land and leave the area, (2) land on the ground and succumb to predation by ants (RIV personal observation), or (3) land and then die of dehydration, starvation, or radiation sickness.

Our findings that holding large numbers of sterile flies in boxes under conditions of high density before release does not affect subsequent fly activity as long as they are held in darkness agree with procedures described by Cunningham *et al.* (1980). Apparently, flies held in darkness remain motionless on the sides of holding boxes, whereas those held in light tend to become overly active under caged conditions.

The advantages of feeding tephritid flies protein are well known. Females fed protein hydrolysate begin laying eggs soon after mating and produce large numbers of eggs (Webster *et al.* 1979, Vargas 1984). Males fed protein exhibit faster development of the reproductive system and changes in adult reproductive behavior (Webster *et al.* 1979). In the present study feeding male flies protein before release appeared to promote greater vigor and movement between leaves within the guava tree canopy. We believe that released sterile male flies that forage actively within the host plant canopy would be in better syncrony with their wild counterparts. We assume that greater residency time within the tree canopy and visits to many leaves are advantageous to eventually finding mates (Baker & van der Valk 1992). Stark *et al.* (1994) studied movement of Oriental fruit fly within and between guava trees in a commercial orchard by knockdown sampling with pyrethrum. Wild males moved actively from leaf to leaf and between trees many times during the day in search of food, water, and mating and resting sites.

We suggest that the methods outlined here could be used by sterile insect program managers to assess the mobility of released sterile flies and to identify superior holding procedures. Our findings further suggest that feeding irradiated flies protein may be advantageous by providing more vigorous sterile males that will forage actively.

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